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2-623

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Art Unit: 1623 1811

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Point of Contact:
Mary Hale
Technical Info. Specialist
CM1 12D16 Tel: 308-4258

Rec'd 3/19

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SERIAL NUMBER	FILING DATE	CLASS	GROUP ART UNIT	ATTORNEY DOCKET NO.
09/086,488	05/28/98	435	1623	ETIP002US

APPLICANT: ROBERT L. JAFFE, LONG ISLAND CITY, NY.

CONTINUING DOMESTIC DATA********
VERIFIED

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371 (NAT'L STAGE) DATA********
VERIFIED

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FOREIGN APPLICATIONS********
VERIFIED

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FOREIGN FILING LICENSE GRANTED 06/11/98 ***** SMALL ENTITY *****

Foreign Priority claimed ☐yes ☐no / 35 USC 119 (a-d) conditions met ☐yes ☐no ☐Met after Allowance / Verified and Acknowledged _____ Examiner's Initials	STATE OR COUNTRY	SHEETS DRAWING	TOTAL CLAIMS	INDEPENDENT CLAIMS
_____ Initials	NY	7	15	1

SEE CUSTOMER NUMBER: 021121

DETERMINATION OF CYTOTOXIC SUBSTANCES IN WHOLE EFFLUENT SAMPLES

FILING FEE RECEIVED	FEES: Authority has been given in Paper No. _____ to charge/credit DEPOSIT ACCOUNT NO. _____ for the following:	☐ All Fees ☐ 1.16 Fees (Filing) ☐ 1.17 Fees (Processing Ext. of time) ☐ 1.18 Fees (Issue) ☐ Other _____ ☐ Credit
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dependent) is the highest number found in the appropriate box in column 1.

CLAIMS

1. A method for evaluating a whole effluent sample for the presence of cytotoxic substances comprising the steps of:

(a) obtaining a sample for testing containing a plurality of potentially cytotoxic substances;

(b) combining a first aliquot of the whole effluent sample directly with a first culture of a particle-feeding flagellate; and

(c) monitoring the growth of the particle-feeding flagellate culture in the presence of the whole effluent sample, wherein a decrease in growth of the culture in the presence of the whole effluent sample is indicative of the presence of cytotoxic agents in the whole effluent sample.

2. The method according to claim 1, wherein the particle-feeding flagellate is *Tetramitus rostratus* in flagellate form.

3. The method according to claim 1, wherein the particle-feeding flagellate is selected from the group consisting of *Chilodenella uncinata, Bodo caudataus, Cercomonas longicauda, Diplonema ambulator, Scytomonas pusilla* and *Bodo designis.*

4. The method according to claim 1, wherein a series of dilutions of the whole effluent sample is prepared and each dilution is individually combined with a culture of particle-feeding flagellate to determine a dose response curve.

5. The method according to claim 4, wherein the particle-feeding flagellate is *Tetramitus rostratus* in flagellate form.

6. The method according to claim 4, wherein the particle-feeding flagellate is selected from the group consisting of *Chilodenella uncinata, Bodo caudataus, Cercomonas longicauda, Diplonema ambulator, Scytomonas pusilla* and *Bodo designis.*

7. The method of claim 1, further comprising the steps of

filtering a second aliquot of the whole effluent sample through a filter having a defined pore size to produce a filtered whole effluent sample from which particulate materials greater in size than the defined pore size have been removed;

combining the filtered whole effluent sample with a second culture of particle-feeding flagellate;

determining the growth of the second particle-feeding flagellate culture in the presence of the filter whole effluent sample; and

comparing the growth of the second particle-feeding flagellate culture in the presence of the filtered whole effluent sample to the growth in the presence of the unfiltered whole effluent sample, wherein as difference in the growth is indicative of the presence of particulate toxic substances in the whole effluent sample.

8. The method of claim 7, wherein a series of dilutions of the filtered whole effluent sample is prepared and each dilution is individually combined with a culture of particle-feeding flagellate to determine a dose response curve.

9. The method according to claim 8, wherein the particle-feeding flagellate is *Tetramitus rostratus* in flagellate form.

10. The method according to claim 7, wherein the particle-feeding flagellate is *Tetramitus rostratus* in flagellate form.

11. The method according to claim 5, further comprising the steps of

recovering a particulate fraction from an aliquot of the whole effluent sample;

combining the particulate fraction with a third culture of particle-feeding flagellate;

determining the growth of the particle-feeding flagellate culture in the presence of the particulate fraction; and

comparing the growth of the particle-feeding flagellate culture in the presence of the particulate fraction to the growth in the presence of the unfiltered whole effluent sample.

12. The method of claim 11, wherein a series of dilutions of the particulate fraction is prepared and each dilution is individually combined with a culture of particle-feeding flagellate to determine a dose response curve.

13. The method according to claim 12, wherein the particle-feeding flagellate is *Tetramitus rostratus* in flagellate form.

14. The method according to claim 11, wherein the particle-feeding flagellate is *Tetramitus rostratus* in flagellate form.

15. The method of claim 1, further comprising the step of monitoring the growth of a second culture of particle-feeding flagellate in the presence of the whole effluent and comparing the growth of the first and second cultures, wherein the mean size of the flagellates in the first and second cultures is different.

Gitomer
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=> s particle feed? flagell? and (unicina? or caudata? or longicaud? or
ambulat? or pusill? or design?)

L1             0 FILE MEDLINE
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L3             0 FILE BIOSIS
L4             0 FILE EMBASE
L5             0 FILE WPIDS

TOTAL FOR ALL FILES
L6             0 PARTICLE FEED? FLAGELL? AND (UNICINA? OR CAUDATA? OR
LONGICAUD?
                 OR AMBULAT? OR PUSILL? OR DESIGN?)

=> s  flagell? and (unicina? or caudata? or longicaud? or ambulat? or pusill?
or designis?)

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L9           313 FILE BIOSIS
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L11            0 FILE WPIDS

TOTAL FOR ALL FILES
L12          335 FLAGELL? AND (UNICINA? OR CAUDATA? OR LONGICAUD? OR AMBULAT?
OR
                 PUSILL? OR DESIGNIS?)

=> s  flagell? and rostrat?

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=> s (l12 or l18) and particle feed?

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L22 1 FILE EMBASE
L23 0 FILE WPIDS

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L24 4 (L12 OR L18) AND PARTICLE FEED?

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PROCESSING COMPLETED FOR L24
L25 1 DUP REM L24 (3 DUPLICATES REMOVED)

=> d cbib abs 1

L25 ANSWER 1 OF 1 MEDLINE DUPLICATE 1
96231558 Document Number: 96231558. Rapid assay of cytotoxicity using Tetramitus **flagellates**. Jaffe R L. (Environmental Toxicology Laboratory, Long Island City, New York 11101, USA.)TOXICOLOGY AND INDUSTRIAL HEALTH, (1995 Sep-Oct) 11 (5) 543-58. Journal code: VWS. ISSN: 0748-2337. Pub. country: United States. Language: English.
AB A simple test for measuring cytotoxic agents has been developed using the **flagellate** phenotype of Tetramitus **rostratus**. The test measures dose-dependent inhibition of cell division by individual agents such as 4-nitroquinoline-N-oxide and other mutagens. Dose-response data are given also for mixtures including coal tar pitch condensate, centrifuged particles obtained from tap water, and water concentrates prepared with XAD-2 resin. Because Tetramitus **flagellates** have a gullet and are **particle-feeders**, the assay allows for cytotoxic measurements of whole particles without prior extraction or solvent substitution procedures. The cytotoxic activities observed may reflect genotoxic activity, since all chemicals that produced a positive response are genotoxic in other test systems.

=> s flagell? and cytotox? and effluent?

L26 0 FILE MEDLINE
L27 0 FILE CAPLUS
L28 1 FILE BIOSIS
L29 0 FILE EMBASE
L30 0 FILE WPIDS

TOTAL FOR ALL FILES
L31 1 FLAGELL? AND CYTOTOX? AND EFFLUENT?

=> d cbib abs

L31 ANSWER 1 OF 1 BIOSIS COPYRIGHT 1999 BIOSIS
1982:201482 Document No.: BA73:61466. LEISHMANIA-ENRIETTII IMMUNE INDUCTION OF MACROPHAGE ACTIVATION IN AN EXPERIMENTAL MODEL OF IMMUNO PROPHYLAXIS IN THE MOUSE. MAUEL J; BEHIN R; LOUIS J. WORLD HEALTH ORGANIZATION IMMUNOL. RES. AND TRAINING CENTRE, INST. BIOCHEM., 1066 EPALINGES, SWITZERLAND.. EXP PARASITOL, (1981) 52 (3), 331-345. CODEN: EXPAAA. ISSN: 0014-4894. Language: English.
AB Some of the parameters of the cellular immune response elicited in mice by inoculation of the nonpathogenic protozoan parasite L. enriettii are described. In vitro incubation of leishmania-infected mouse peritoneal macrophages with spleen cells from syngeneic Leishmania-immune animals

resulted in activation of the paghocytes, leading to intracellular parasite destruction. Activation required interaction of sensitized lymphocytes with parasite antigen released or displayed infected macrophages. The effect was dependent both on the dose of parasites used for in vivo priming and on the number of spleen cells cocultivated with parasitized by anti-Thy-1 antiserum treatment and was retained in the **effluent** cells after nylon-wool separation. Activation was followed by lysis of part of the macrophage monlayer. Destruction of the phagocytes did not appear to result from the activation process per se and may represent a **cytotoxic** activity of sensitized lymphocytes for macrophages bearing parasite antigen on their surface.

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=> s jaffe r?/au,in

'IN' IS NOT A VALID FIELD CODE
L32         658 FILE MEDLINE
L33         627 FILE CAPLUS
L34         764 FILE BIOSIS
'IN' IS NOT A VALID FIELD CODE
L35         536 FILE EMBASE
L36          17 FILE WPIDS

TOTAL FOR ALL FILES
L37        2602 JAFFE R?/AU,IN

=> s l37 and flagell?

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TOTAL FOR ALL FILES
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=> s l43 not (l31 or l24)

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L48           1 FILE WPIDS

TOTAL FOR ALL FILES
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PROCESSING COMPLETED FOR L49
L50            1 DUP REM L49 (1 DUPLICATE REMOVED)

=> d cbib abs


L50  ANSWER 1 OF 1  CAPLUS  COPYRIGHT 1999 ACS          DUPLICATE 1
1995:420587   Document No. 122:180888  Detection of cytotoxic agents using
     Tetramitus rostratus.  Jaffe, Robert L. (USA).  U.S. US 5387508
     A  19950207, 17 pp.  (English).  CODEN: USXXAM.  APPLICATION: US
92-883257
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19920514.
AB Cytotoxic agents, and particularly DNA-damaging agents, can be detected in a sample by a method comprising the steps of (a) adding the sample to a living culture of Tetramitus rostratus in **flagellate** form, (b) detg. the growth rate of the T. rostratus culture in the presence of the sample, and (c) comparing the growth rate of the T. rostratus culture in the presence of the sample to a std. growth rate. A decrease in growth rate is indicative of the presence of cytotoxic agents in the sample. The use of the **flagellate** T. rostratus allows this assay to be used on solid as well as liq. or gaseous samples because T. rostratus ingests particulate materials via a gullet.

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=> s (l12 or l18) and cytotox?

L51             1 FILE MEDLINE
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L53             1 FILE BIOSIS
L54             1 FILE EMBASE
L55             1 FILE WPIDS

TOTAL FOR ALL FILES
L56             6 (L12 OR L18) AND CYTOTOX?

=> s l56 not (l31 or l24 or l43)

L57             0 FILE MEDLINE
L58             0 FILE CAPLUS
L59             0 FILE BIOSIS
L60             0 FILE EMBASE
L61             0 FILE WPIDS

TOTAL FOR ALL FILES
L62             0 L56 NOT (L31 OR L24 OR L43)

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